

## Review Article

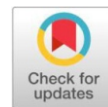
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# Comprehensive review on bacterial wilt of solanaceous crops: Pathogenesis, impact, and management strategies

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## ABSTRACT

Bacterial wilt of solanaceous crops is caused predominantly by *Ralstonia solanacearum* (Smith) and is a soil-borne pathogen. The bacterium *R. solanacearum* is gram-negative, small rod with rounded ends, non-fluorescent, non-endospore forming, strict aerobe and has polar flagella (when present). All phylotypes i.e. I, IIA, IIB, III & IV, race 1 & 3 and biovars 1, 2, 2T, 3 & 4 infect solanaceous crops resulting wilt disease. The pathogen has variability during its survival in the soil. This pathogen is diverse, containing several races, biovars, and phylotypes, which hinders diagnosis and control strategies. It survives in the soil, water, crop residues, and weed hosts for long periods, and therefore, elimination is difficult. Its broad host range combined with the latent infection of plants prevent early detection, and visible symptoms are generally observed when the crop is significantly infected. The study looks at the complex nature of the disease and reviews integrated methods for controlling it. A holistic approach includes microbes, organic matter, botanicals, animal wastes, and resistant varieties which are used either alone or combined with other methods to improve disease management. The microbes particularly fungi and bacteria have an advantage over chemical control such as self-sustaining, self-spreading, reduced input by renewal capacity, long-term disease suppression, and eco-friendly. The research offers important insights into the pathogen's biology. It reveals how it survives, its broad host range, and its genetic diversity. These factors help the pathogen persist and adapt in different agro-ecosystems. By evaluating various control strategies, including cultural, chemical, biological, and host resistance methods, the study identifies the strengths and weaknesses of each approach in real-world conditions. The research particularly emphasizes using biological control agents and combining them with chemical and cultural practices. This integrated approach showed better effectiveness than using any one method alone. Furthermore, the study helped develop practical techniques for disease surveillance. It underlined the need for easy-to-use diagnostic tools and awareness programs for farmers. Overall, this work not only enhances scientific knowledge but also offers a blueprint for practical application in the field. The goal is to reduce yield losses and promote sustainable production of solanaceous crops in areas affected by bacterial wilt. The primary aim of this review is to collate and analyze current knowledge on bacterial wilt, with a particular focus on its impact on solanaceous crops and the efficacy of various management strategies.

**Keywords:** Bacterial wilt, Bioagents, Biocontrol, Integrated disease management, Biovar, Solanaceous crop.

## INTRODUCTION

Nightshade crops or solanaceous crop including potatoes (*Solanum tuberosum* L.), tomatoes (*S. lycopersicum*), eggplant (*S. melongena* L), capsicum and chilli are rich source of vitamins, minerals, phenols, anthocyanins and amide proteins and are called as protective foods. Tomatoes, Potato tubers contain a moderate amount of vitamin C, antioxidants observed in potato many phytochemicals such as lycopene (60 - 64%, phytoene (10 - 12% carotenoids (10-15%) neurosporene (7- 9%) (Clinton 1998). Tomatoes and their products are the richest sources of lycopene in vegetables and also a good source of vitamin A. Potatoes have proteins of high quality having plentiful metabolites that help enhance protein utilization-include  $\alpha$ -tocopherol (0.5 to 2.8 mg/kg), lutein (0.13 to 0.6 mg/kg), and  $\beta$ -carotene (1 mg/kg) (Lachman *et al.*, 2000). Capsicum is a good source of vitamins C, K, carotenoids, and flavonoids. *Capasicum* have high levels of

vitamins and minerals and provide in % of recommended daily allowance, 240% of ascorbic acid, 39% of vitamin B6 (pyridoxine), 32% of vitamin A, 13% of iron, 14% of copper, 7% of potassium in 100 g of produce (Szallasi and Blumberg, 1999; Frei and Lawson, 2008). Folic acid and vitamins can help in the treatment of arthritis and asthmatic problems. Eggplant contains important phytochemicals comprising phenolic compounds (caffeic and chlorogenic acid), and flavonoids, (nasunin). Nasunin or delphinidin-3-(coumaroyl)rutinoside 5-glucoside is the major phytochemical in brinjal (Jorge *et al.*, 1998). It is found highly efficacious in controlling high blood cholesterol.

Solanaceous vegetable crops suffer from biotic and abiotic factors which significantly reduce the production and productivity of the crops worldwide. In biotic factors, diseases caused by different fungi, bacteria, phytoplasma, viruses, viroids and nematodes play a major role in the reduction of crop yields. In these vegetable crops, the bacterial wilt caused by the *Ralstonia solanacearum* (Smith), is one of the major diseases, that occur in the wet tropics, subtropics and some temperate climatic areas of the world (Yabuuchi *et al.*, 1995). The pathogen has international importance due to its A2 quarantine status affecting a very wide range of crops including 450 plant species

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and 54 botanical families. In addition, race 3 biovar 2 has been listed as a select agent in the Agriculture Bioterrorism Act of 2002. The losses caused by bacterial wilt in solanaceous crops varied from 20-100% (Mishra *et al.*, 1995; Wang and Lin, 2005; Singh *et al.*, 2010; Muthoni *et al.*, 2012; Ramesh *et al.*, 2014) world over depending on the host cultivars, environmental conditions and virulence of the pathogen. In India, the solanaceous vegetable crop during the summer season is less affected by the disease in the plains where the temperature is high, but in hills, term disease suppression and eco-friendly In the rainy season, tomato, potato, brinjal and chilli are affected by *R. Solanacearum* in plains, coastal hilly areas and cause losses in 20 - 100 % chilli 2- 60% in tomatoes (Singh *et al.*, 2010) and about 37% in potato and 30 - 100 % in brinjal (Ramesh *et al.*, 2014)

India has faced significant challenges with bacterial wilt in potato crops, particularly in states like Uttar Pradesh and Punjab. Outbreaks of bacterial wilt have led to severe yield losses and increased input costs for potato farmers (Kumar *et al.*, 2014). The economic impact has been substantial, with affected farmers experiencing financial hardship. Efforts to control the disease have included public awareness campaigns and government-supported research into resistant varieties (Singh *et al.*, 2016). This case study in Tamilnadu examined the combined effect of soil solarization and organic amendments on bacterial wilt in eggplant fields. The results showed a substantial reduction in disease incidence and improved soil health, promoting sustainable agricultural practice.

### Pathogen Description: *Ralstonia solanacearum*

#### Taxonomy and Morphology

*Ralstonia solanacearum* is a soilborne, gram-negative bacterium that belongs to the class Betaproteobacteria, order Burkholderiales, and family Burkholderiaceae. It was initially described as *Pseudomonas solanacearum* before being reclassified into the genus *Ralstonia* based on phylogenetic studies.

**Morphology:** The bacterium is rod-shaped, measuring about 0.5-0.7 µm in diameter and 1.5-2.5 µm in length. It is motile, equipped with a single polar flagellum, and can form biofilms, enhancing its survival and infectivity.

**Colony Characteristics:** On a tetrazolium chloride (TZC) agar medium, colonies of *R. solanacearum* are typically fluidal, irregular, and white with a pink centre.

**Genomic Insights:** The genome of *Ralstoniasolanacearum* is complex and large, comprising two circular chromosomes and a megaplasmid. The genomic content varies among strains, contributing to the pathogen's adaptability and broad host range.

- **Genome Size:** The genome size ranges between 5.8 and 6.1 Mb, with around 5,000-5,500 predicted protein-coding genes.
- **Virulence Genes:** Key virulence factors include type III secretion system (T3SS), type IV pili, exopolysaccharide (EPS) production, and plant cell wall-degrading enzymes.

#### Pathogen Variability and Diversity

*R. solanacearum* is known for its genetic diversity, which is reflected in its classification into races, biovars, and phylotypes based on host range, biochemical properties, and phylogenetic relationships.

#### Races and Biovars:

- **Races** are determined by host range. For example, Race 1 affects a wide range of plants, Race 2 primarily infects bananas, and Race 3 is mainly found in potatoes.
- **Biovars** are classified based on the ability to utilize different carbon sources, with biovars 1, 2, 3, 4, and 5 recognized.

**Phylotypes:** A more recent classification divides *R. solanacearum* into four phylotypes based on phylogenetic analysis:

- Phylotype I: Asian origin
- Phylotype II: American origin
- Phylotype III: African origin
- Phylotype IV: Indonesian origin

#### Host Range and Symptomatology

*R. solanacearum* has a broad host range, infecting over 200 plant species across 50 families. Solanaceous crops such as tomato, potato, eggplant, and pepper are particularly susceptible.

#### • Symptoms:

- **Wilting:** The most characteristic symptom is wilting of the plant during the hottest part of the day, which may recover overnight in the early stages.
- **Stunting and Yellowing:** Infected plants exhibit stunted growth, yellowing of leaves, and eventual necrosis.
- **Vascular Discoloration:** Brown discoloration of the vascular tissue is a key diagnostic feature. Cutting the stem reveals a slimy exudate.

#### Epidemiology and Disease Development

The epidemiology of bacterial wilt is influenced by environmental factors, modes of transmission, and pathogen survival strategies.

*Xanthomonas campestris*, in contrast, is less genetically diverse but still poses a significant threat due to its specific host range and ability to cause severe symptoms (Poussieret *et al.*, 2003). This bacterium is characterized by the formation of water-soaked lesions and can cause systemic infection, leading to plant death. Bacterium is introduced into the soil through contaminated water or plant debris. It enters the plant through root wounds or natural openings and colonizes the xylem vessels: Once inside the xylem, *Xanthomonas campestris* proliferates and obstructs water and nutrient transport, causing the characteristic wilting symptoms. The bacterium can spread systemically through the plant, leading to severe damage. Like *Ralstonia solanacearum*, *Pseudomonas solanacearum* infects through the root system and moves through the xylem vessels. It produces a range of toxins and extracellular polysaccharides that contribute to its ability to cause disease. Infection Process bacterium enters the plant through wounds or natural openings, colonizes the xylem, and produces extracellular polysaccharides that block water flow. Virulence Factors secretes various exopolysaccharides and has a unique set of effector proteins that contribute to its pathogenicity.

*Pseudomonas solanacearum* is less adaptable than *Ralstonia solanacearum* but still thrives in warm and moist conditions. It is less commonly found in extreme tropical climates but can be problematic in subtropical regions. In addition to *Ralstonia solanacearum* and *Pseudomonas solanacearum*, several other bacteria can cause wilt diseases in solanaceous crops although they are less prevalent.

**Corynebacterium Species:** These bacteria can cause wilt-like symptoms but are less common and typically occur in specific conditions or regions.

**Xanthomonas Species:** Some species within this genus can cause wilting, although their impact is generally limited compared to *Ralstonia solanacearum*. The bacterium survives in soil, water, and plant debris. It can form biofilms and persists in the soil for an extended period. The bacterium infects through roots, particularly through wounds or natural openings. Once inside the plant, it spreads through the xylem vessels, causing systemic symptoms. The pathogen can be dispersed by water, soil, and contaminated tools or plant material. Several environmental factors influence the survival, virulence, and spread of bacterial wilt pathogens. Both *Ralstonia solanacearum* and *Pseudomonas solanacearum* are more virulent at higher temperatures, typically between 25°C and 30°C. Soil Moisture: High soil moisture levels promote the survival and spread of these pathogens. Conversely, dry conditions can limit their spread. Soil pH and Composition: affect pathogen survival and plant presence of susceptible host plants can influence the prevalence of bacterial wilt.

### Pathogenic Mechanisms

***Ralstonia solanacearum*** infects plants through the roots, where it enters the vascular system. The bacterium produces a range of virulence factors, including extracellular polysaccharides (EPS), which form biofilms and protect the bacterium from environmental stressors. It also secretes various effector proteins that manipulate plant host defenses, leading to wilting and ultimately plant death. The infection begins when the bacterium is introduced into the soil through contaminated water or plant debris. It enters the plant through root wounds or natural openings and colonizes the xylem vessels: Once inside the xylem, *Ralstonia solanacearum* proliferates and obstructs water and nutrient transport, causing the characteristic wilting symptoms. The bacterium can spread systemically through the plant, leading to severe damage. *Ralstonia solanacearum* thrives in warm, moist conditions. It has a wide temperature range for growth, but it is most virulent at temperatures between 25°C and 30°C. The bacterium can survive in the soil for extended periods, especially in regions with high humidity and organic matter. It prefers well-drained soils and can persist in both tropical and subtropical climates. Soil moisture and pH levels also influence its survival and infectivity.

***Pseudomonas solanacearum***, formerly classified under the genus Burkholderia, has been reclassified into the genus Pseudomonas. It is a gram-negative bacterium and is part of the Pseudomonadaceae family. This pathogen is less common but still notable for causing bacterial wilt in specific environments. *Pseudomonas solanacearum* has a smaller genome compared to *Ralstonia solanacearum*, with fewer pathogenicity islands. The genetic diversity within this species contributes to its varying levels of virulence and host range. Like *Ralstonia solanacearum*, *Pseudomonas solanacearum* infects through the root system and moves through the xylem vessels. It produces a range of toxins and extracellular polysaccharides that contribute to its ability to cause disease. Infection Process bacterium enters the plant through wounds or natural openings, colonizes the xylem, and produces extracellular polysaccharides that block water flow. Virulence factors secrete various exopolysaccharides and has a

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### Management Challenge

Recent advances in research have provided new tools and strategies for managing bacterial wilt. Molecular techniques for pathogen detection and identification have improved our understanding of pathogen diversity and distribution, enabling more targeted management approaches (Mansfield *et al.*, 2012). Genetic research has led to the development of resistant crop varieties, offering a promising solution to reduce the impact of the disease (Fegan & Prior, 2005). Innovations in biological control, such as the use of antagonistic microorganisms and biofungicides, have shown potential in managing bacterial wilt (Sikora *et al.*, 2020). These methods offer sustainable alternatives to chemical control and contribute to integrated management strategies. Future research should focus on enhancing disease resistance, improving diagnostic tools, and developing sustainable management practices. The integration of new technologies and approaches will be crucial for addressing the ongoing challenges associated with bacterial wilt and improving crop health and productivity.

Bacterial wilt, caused primarily by the soil-borne bacterium *Ralstonia solanacearum*, is a major threat to solanaceous crops. Other pathogens, including *Pseudomonas solanacearum* and various less common species, also contribute to the disease. This section provides a detailed examination of these pathogens. Lesions can restrict the international trade of infected crops, leading to economic losses in exporting countries (Lal *et al.*, 2020)

Cultural practices are foundational to managing bacterial wilt and involve modifications to farming techniques and crop management. Growing resistant crop varieties is one of the most effective ways to manage bacterial wilt.

Breeding programs have developed several resistant varieties, using commercially available resistant varieties of tomatoes, potatoes, and eggplants. For example, in varieties like 'Solar Set' for tomatoes and 'Patriot' for potatoes proper sanitation reduces the spread of bacterial wilt by removing infected plant debris. Controlling vectors regularly remove and destroy infected plants. Clean and disinfect tools and equipment used in the field (Glick, 2012). Use 1-2% sodium hypochlorite solution to disinfect tools and equipment. Ensure thorough cleaning to reduce pathogen transfer.

Chemical control methods involve the application of bactericides and soil treatments to manage bacterial wilt. Use copper-based bactericides such as copper hydroxide or copper oxychloride. These are effective in reducing pathogen populations (Morris et al., 2017). Apply Copper hydroxide at 2-3 kg per hectare, mixed with water according to the manufacturer's recommendations. Apply as a foliar spray by Integrated Approach. Combining bactericide applications with crop rotation and resistant varieties for improved control. Monitor and adjust based on disease pressure (Van den Berg et al., 2017).

Certain bacteria can suppress the growth of *Ralstonia solanacearum* and reduce disease severity. As for example, using strains of *Bacillus subtilis*, *Pseudomonas fluorescens*, or *Streptomyces* species. These bacteria can produce antimicrobial compounds that inhibit pathogen growth (Kloepper et al., 2004). *Bacillus subtilis* applying at  $1-2 \times 10^8$  CFU per hectare, mix with water and apply as a soil drench or foliar spray every 2-3 weeks during the growing season. (Kloepper, J. W., et al., 2004). "Biological control of bacterial mycorrhizal fungi (AMF) such as *Glomus* spp. @10-20 kg per hectare, improve plant AMF Inoculants, nutrient uptake and can suppress pathogen activity (Smith & Read, 2008). Incorporate into the soil before planting or apply as a soil drench with using antagonistic bacteria.

Advancements in genomics have provided significant insights into the biology and variability of bacterial pathogens. *Ralstonia solanacearum*, the primary agent of bacterial wilt, exhibits considerable genetic diversity, which complicates disease management (Fegan & Prior, 2005). Recent genomic studies have identified key virulence factors and mechanisms of pathogenesis. For instance, the complete genome sequencing of various strains of *Ralstonia solanacearum* has revealed differences in the pathogenicity islands and effector proteins, which are crucial for host interaction (Cohn et al., 2014).

The availability of genome sequences has enabled researchers to develop more precise molecular markers for strain identification and tracking. This information is essential for understanding the epidemiology of bacterial wilt and for developing targeted control measures (Wang et al., 2016). Additionally, comparative genomics has highlighted potential targets for novel therapeutic strategies and provided insights into the evolution of the pathogen (Hao et al., 2021).

Understanding the interactions between *Ralstonia solanacearum* and its host plants is critical for developing resistant varieties. Recent advances in molecular biology have shed light on the mechanisms through which the pathogen evades host defenses and establishes infection. Studies on the plant immune system, including the roles of pattern recognition receptors (PRRs) and resistance (R) genes, have enhanced our understanding of host-pathogen dynamics (Jones & Dangl, 2006).

Research has focused on identifying and characterizing plant genes involved in resistance to bacterial wilt.

For example, the identification of resistance genes in tomatoes, such as the *I* gene and its derivatives, has paved the way for marker-assisted selection in breeding programs (Maroof et al., 2017). These advances in genomics and molecular biology are crucial for developing crops with durable resistance to bacterial wilt. Genetic engineering techniques, including CRISPR/Cas9 gene editing, have revolutionized plant research and offer new avenues for managing bacterial wilt. The CRISPR/Cas9 system allows for the precise modification of plant genomes, enabling the introduction of resistance genes or the knock-out of susceptibility genes (Mali et al., 2013). Recent studies have applied CRISPR technology to develop solanaceous crops with enhanced resistance to *Ralstonia solanacearum* by targeting specific susceptibility genes (Zhu et al., 2020).

Moreover, genetic engineering has facilitated the development of transgenic plants expressing antimicrobial peptides or proteins that inhibit bacterial growth. These approaches have shown promise in laboratory and greenhouse trials, although further research is needed to assess their effectiveness in field conditions (Gao et al., 2016). Early and accurate detection of bacterial wilt is crucial for effective disease management. Recent advances in molecular diagnostic techniques, such as polymerase chain reaction (PCR) and real-time PCR, have significantly improved the ability to detect *Ralstonia solanacearum* and other bacterial wilt pathogens (Pang et al., 2016). These techniques offer high sensitivity and specificity, allowing for the detection of low pathogen concentrations in soil, plant tissues, and irrigation water (Li et al., 2018).

Additionally, loop-mediated isothermal amplification (LAMP) has emerged as a rapid and cost-effective diagnostic tool. LAMP assays can be performed without sophisticated equipment and provide results in less than an hour, making them suitable for on-site detection in field conditions (Notomi et al., 2000). Recent studies have optimized LAMP assays for the detection of *Ralstonia solanacearum*, enhancing their utility for rapid disease monitoring and management (Wang et al., 2020). This section has provided a comprehensive overview of the primary bacterial wilt pathogens affecting solanaceous crops, including their taxonomy, genetic characteristics, pathogenic mechanisms, and environmental interactions. Understanding these aspects is crucial for developing effective management strategies to combat bacterial wilt and mitigate its impact on agriculture.

The epidemiology of bacterial wilt is influenced by environmental factors, modes of transmission, and pathogen survival strategies.

- **Environmental Factors:** The disease is favored by warm (25-35°C), humid conditions and high soil moisture. It is prevalent in tropical and subtropical regions but can also occur in temperate zones under suitable conditions
- **Transmission:** The pathogen is primarily soilborne but can also be spread through contaminated irrigation water, infected plant material, and agricultural tools. Insects like the root-knot nematode (*Meloidogyne* spp.) can also facilitate its spread.
- **Survival:** *R. solanacearum* survives in soil, water, and plant debris. It can persist in the environment for extended periods, forming biofilms and entering a viable but non-culturable (VBNC) state under adverse conditions.

## Conclusion

Future research directions in bacterial wilt management encompass a wide array of innovative and interdisciplinary

approaches, from advanced genomics and resistant variety development to integrated disease management and precision agriculture. These efforts aim to develop sustainable, effective, and adaptable strategies to combat *Ralstonia solanacearum* and mitigate the impact of bacterial wilt on solanaceous crops. These perspectives and research directions, supported by relevant references, provide a comprehensive outlook on the future of bacterial wilt management. They highlight the need for continued research, collaboration, and innovation to address this persistent agricultural challenge.

### Future Scope of Study

The future plan for studying bacterial wilt in solanaceous crops like tomato, brinjal, and chilli focuses on a thorough approach. This plan looks at pathogen variability, host resistance, disease ecology, and management strategies. The first phase will involve isolating and characterizing *Ralstonia solanacearum* strains from major crop-growing areas. We will use molecular tools, including PCR-based assays, Rep-PCR, and multilocus sequence analysis (MLSA), to explore genetic diversity, virulence patterns, and race/biovar classification. Next, we will conduct large-scale disease surveillance. This includes seasonal field surveys and geo-tagging to identify disease hotspots. We will correlate disease incidence with soil and climatic conditions, aiming to develop prediction models. At the same time, we will screen available germplasm, landraces, wild relatives, and released cultivars. Our goal is to find sources of resistance through phenotypic evaluations, both in controlled conditions and natural hotspots. This data will be used in breeding programs to create resistant lines. To manage the disease sustainably, we will create and evaluate integrated disease management (IDM) modules. These will include biocontrol agents like *Bacillus subtilis*, *Pseudomonas fluorescens*, and *Trichoderma* spp. We will also use soil amendments, including compost, vermicompost, neem cake, and plant extracts with antibacterial properties. Additionally, we will explore the application of metal and polymer-based nanoparticles for pathogen control. We will consider the potential use of CRISPR/Cas systems for gene editing to develop resistance. We will analyze soil microbiomes using next-generation sequencing to identify and improve beneficial microbial populations in soils that suppress disease. Furthermore, we will optimize grafting techniques using resistant rootstocks and crop rotation practices to reduce disease pressure. Alongside these laboratory and field interventions, we will focus on capacity-building and knowledge transfer. This will involve farmer field schools, mobile advisory platforms, participatory demonstrations, and training for extension personnel to promote the adoption of effective practices. We will seek collaboration with ICAR institutes, state agricultural universities, Krishi Vigyan Kendras (KVKs), and international plant pathology research centers. This will help strengthen data sharing, conduct joint field trials, and quickly scale up validated technologies. Overall, this integrated plan aims to create a long-term, farmer-friendly strategy to reduce the damaging impact of bacterial wilt on the productivity of solanaceous crops and farmers' income.

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